

Learning From Patients: The Science of Medicine (2003)
Lecture One—Research Mechanics: Putting the Brakes on Cancer
Bert Vogelstein, M.D.

1. Start of Lecture 1 (00:11)

From the Howard Hughes Medical Institute the 2003 Holiday Lectures on Science. This year's lectures Learning from Patients: The Science of Medicine will be given by Dr. Bert Vogelstein Howard Hughes Medical Institute Investigator at Johns Hopkins University School of Medicine and Dr. Huda Zoghbi Howard Hughes Medical Institute Investigator at Baylor College of Medicine. The first lecture is titled Research Mechanics: Putting the Brakes on Cancer. And now, to introduce our program the president of the Howard Hughes Medical Institute Dr. Thomas Cech.

2. Introduction by HHMI President Dr. Thomas Cech (01:00)

Welcome to the Howard Hughes Medical Institute and to our eleventh annual Holiday Lectures on Science. This program is being webcast live and in addition we have almost two hundred students in the audience from the Greater Washington, D.C. area high schools. This series is going to be available on DVD starting in the spring of 2004 and you can order these DVDs through our web site. Now, the web site is also a great place to learn about all of the educational programs supported by the Howard Hughes Medical Institute and, in addition, about the research activities of our 323 Howard Hughes Medical Institute investigators who run laboratories across the United States. This particular series is called Learning from Patients: The Science of Medicine and this is really a two-way street. We're going to talk about the way that biomedical scientists derive the inspiration for their research from the patients that they see in the clinic but then, in the other direction after figuring out the basic details of the disease process they try to turn around and find ways that they can use this understanding to improve diagnosis and hopefully, eventually, treatment of these diseases. Now, for this exploration we couldn't have better guides than our two speakers for this series. They are both MDs. They both continue to have contact with patients while they're doing their cutting-edge research. Huda Zoghbi is interested in patients that have problems with their nervous system. You'll hear about that tomorrow. Today, we have Bert Vogelstein who's going to be talking about genes that are involved in the process of cancer and also particular interest in translational research. This is a term that is used to describe trying to translate or apply, the information that's learned in the research laboratory back into the clinic. Now, Bert's going to talk for forty minutes. He will ask for questions halfway through the lecture and then again at the end so you can already be thinking as he's talking about what you might ask. The title of his talk is Research Mechanics: Putting the Brakes on Cancer.

3. Introductory interview with Dr. Bert Vogelstein (03:42)

I did a residency in pediatrics. That's when I first met my first cancer patients. That made a large impact on me. At that point, no one had any real idea what cancer was. It just seemed like some mysterious disease that affected people it could come from outer space, as far as we knew and that was really very hard to be hopeful with the parents or with the kids. So at that point, I think my goal solidified to doing research on cancer and I did my postdoctoral fellowship in a cancer research laboratory and I've been doing cancer research ever since that time. There's been a huge change in terms of understanding of cancer I mean, a real revolution. There's still lots of details to be learned and some major things still to be learned but a general outline of how cancers form has been generated and you can kind of summarize that revolution in a single sentence. The sentence would be "Cancer is, in essence, a genetic disease." So from pessimism or at least a feeling of hopelessness in the seventies has turned to real optimism in the new century because once you understand a disease history shows that it's only a matter of time before

you can do something about the disease and what's critical for the future is to get young people with creative ideas to work on this problem and to solve it in their lifetime and I think that's really possible now. I hope my lectures will accomplish a couple of things to educate the students about what cancer is. I want to convey to them some of the excitement that's now widely felt in the scientific community about cancer and I want to stimulate those of them who are thinking about science as a career to think harder about it, and to understand that this is a field where they really can make an impact a difference on millions of people worldwide. It's just a wonderful way to spend your life.

4. Outline of the lectures (06:19)

Well, thank you. I'd also like to welcome you here to Howard Hughes Medical Institute. You are an essential part of this program. There will be hundreds of thousands of high-school students who view these lectures in the coming years but you have the advantage in that you're the only ones allowed to ask questions to us because the rest will only see the CDs. So you represent all of the other high-school students and please think about what questions all high-school students may have, including those that you might have and as Dennis said before we have these wonderful lights. Just hold up a light when you want to ask a question. We're going to reward some questioners with T-shirts so look out. I'm also going to ask you some questions as we go along. I'm going to try to make this an interactive program. I don't know many of your names but I'll just pick random names and hope that strikes a bell. Any of you named Jim or James? That's good. How about Anne or Anna? Anybody named Mauricio? Got it. Nirika? OK. I think we got all the common names covered, then. So let's start with a patient that was mentioned in that tape Melissa. She was my first patient. This was really tragic not only because Melissa was such a beautiful little girl and because she developed cancer but also because at that time which was in the seventies we really knew very little about this disease. It seemed just to be something which came from outer space and it was so hard to be optimistic that we would ever be able to do anything for kids like Melissa or older patients with cancer until we understood something. Now, thirty years later, we do understand something. There's been a real revolution in cancer research and I'm going to try to introduce you to that revolution in these two lectures. There will be four chapters. The first chapter will be the nature of cancer what is cancer? The second will be an explanation of the genes that cause cancer what they are and what they do. The third will be focused on how we use that information to prevent cancer in the future and the fourth will be how we might be able to use that information to treat cancers again, in the future...

5. Nature of cancer: Malignancy and metastasis (09:15) and let's begin with the nature of cancer.

There are lots of different types of tumors and I'll tell you the difference between cancer and tumors in a second but cancers are a form of tumor. This is another form of a tumor but it's benign, not malignant, as cancers are. Benign tumors are not life-threatening. They're not particularly dangerous. You can see that small tumor on this young woman's forehead. You might call it a birthmark. Probably all of you have one or two of these. Scientifically, they're called nevi or nevus and they're really not very important. Some people even think they are beauty marks. Another kind of tumor is shown here on the vocal cords. There is two of them. These are also benign. They're not dangerous. They probably wouldn't cause these patients any problems unless the patient was Britney Spears or Michael Jackson but those people have more important problems to worry about than their voice, at present. Tumors are not benign tumors are not defined by their size. I showed you ones that are small but here is a slightly bigger one growing out of the salivary gland of this gentleman's face. This gentleman was lived was a Native American lived on a reservation and, in fact, wasn't able to get to a hospital for a long time. It took about thirty years to grow a tumor this size but it's still benign so when he eventually reached the hospital a surgeon was able to cut out the tumor and the patient went home and lived another thirty years.

6. Nature of cancer: Malignancy and metastasis (11:16)

So it's not size that defines the difference between a benign tumor and a malignant tumor. Another name for a malignant tumor is a cancer. It's not size. What is it? It's something called invasiveness. On your left is another birthmark a nevus. It's a tumor of pigment cells in your skin that determine the color of your skin and the benign tumor is round. It's circumscribed. It has smooth borders. It can be removed easily by a dermatologist or a surgeon but the one on the right is malignant and the difference is not so much the size but the fact that it's not smooth any longer. It's invading. It's invading through the skin laterally and more importantly it's also invading in three dimensions so it's invading underneath the skin. It will eventually hit a blood vessel or a lymphatic and then tumor cells can go inside the vasculature that is, the vessels, the blood vessels and travel to other parts of the body. That is called metastasis when a malignant tumor reaches a blood vessel or lymphatic travels throughout the bloodstream reaches other organs like the lungs or the liver and starts to form a new tumor a kind of colony of the original tumor in the new place and that's why people die, because of metastases. The primary tumors can almost always be removed surgically but if the tumors aren't detected until after they have metastasized then it's too late. The Greeks recognized this invasiveness of cancer very early on, and that's why they called it "cancer" which means "a crab." They saw the invasive arms of the cancer such as in the melanoma I just showed you and that reminded them of the claws of a crab,

7. Animation: Tumor growth and metastasis (13:21)

and this animation shows what happens in a tumor. There are cells which are dividing abnormally. We'll see how in a few minutes. They keep dividing at the expense of the normal cells that surround them and in three dimensions, once they divide enough they start to pop out. You can begin to see them with your naked eye not just under a microscope and then they recruit blood vessels. They need to recruit blood vessels in order to grow to a size that's greater than just a tenth of an inch in diameter. The blood vessels supply oxygen and nutrients and the last stage is the metastatic stage. That blue cell in the middle represents a metastatic cell that has come from the primary tumor invaded into the blood vessel and now can also get out of the blood vessel invade another tissue and form new cells there.

8. Examples of metastatic cancer (14:32)

The end process of the metastatic lesion the metastatic process, is shown here. On your left is a normal lung, an x-ray and on the right is a picture of a patient who originally had a breast tumor that had metastasized to the lung. So the tumor cells from the breast had invaded a blood vessel and traveled to the lung where they have formed nodules composed of breast cancer cells. Another example is provided by colon cancers. Colon cancers start in the colon, of course and they invade a different kind of vessel. It's called a portal vein and it connects the intestines to the liver. The cells which invade wind up in the liver and on this next slide you can see a normal liver on your left and on the right is a liver that has been invaded by colon cancer cells metastatic colon cancer cells. Each of those nodules represents an individual cancer cell which has made its way to the liver and started to multiply. Each of those nodules now contains several hundred million cells and the liver will eventually be totally replaced by cancer. A person can't live without his liver so that's why this person will die.

9. Many types of cancer (16:04)

Now, there are lots of kinds of cancers. Cancer is often thought of as a single disease in the newspaper but it's really not. It's hundreds of different diseases. For every person in this room, there's at least one form of cancer. For every cell type in the body there's at least one form of cancer and they're different. For example, a breast cancer is different than a melanoma or a colon cancer. On your left is a normal breast a mammogram the kind of test that women are expected to get so that breast tumors can be detected before they're metastatic and on your right is an example of a breast tumor a cancer, that has

been detected by mammography. Another kind of cancer that particularly affects young people is leukemias. On your left is a normal blood smear. That is a drop of blood that's taken from a patient and smeared on a slide really, just smeared and then stained with dyes. You can see all the red cells. Most of the cells in the blood are, of course, red cells but there are also a few white cells. Those are normally there and they help prevent infections guard against infections but in a blood smear from a leukemia patient there are still a few normal-looking white cells but most of the white cells are these large and abnormal forms which represent the leukemia cells and because these cells are already in the blood they're a cancer of blood cells they spread immediately throughout the body and they have to be treated immediately for the patient to survive.

10. Tumors and ratio of cell birth to death (17:48)

Now, despite the fact that all of these tumors are different in some ways they also have something in common and what they have in common is that there is an abnormal ratio between cell birth and cell death. So in a normal adult tissue say, the skin or in the intestines cells are always dividing in the skin or in the intestines there's always a very specific ratio of cell birth and cell death. Can anybody tell me what that ratio is? Just raise your hand or your lighter. One. Exactly. Has to be exactly one. For every cell that's born, one cell dies. If too many cells die more cells die than are born the tissue will shrink. It will atrophy. On the other hand, if more cells are born than die that's a tumor. That's the physiologic definition of a tumor and it doesn't require much of an increase of that ratio in order to get a big tumor over time.

11. Compound-interest analogy (19:01)

You can think about this like interest. Any of you working in a lab this summer? OK. So if I were to ask you if I were to offer you a million dollars to work in my lab over the summer you would probably take it, right? OK. I'm not going to offer you that much. Suppose I offer you a penny, but I tell you that every day you'll get twenty-five percent interest on that penny. So after one day, you'll have 1.26 cents and after two days, you'll have 1.6 cents and three days, 1.9 cents. Would you rather have the penny with interest or the million dollars? Everybody, who would rather have the penny? - How long? - One hundred days. One summer. And who would rather have the million dollars? OK. Good. So most of you chose wisely. A few of you chose poorly. Those of you who took the penny would end up with \$49 million. Those of you who took the million you wouldn't do so bad, but you'd still only have a million after the end of the summer. So now, this same kind of thing happens in tumors. It only requires a slight difference in the ratio of cell birth to cell death to develop a very large tumor over time and since most tumors take twenty to thirty years to develop it really only requires a difference of one percent in that ratio to get a huge tumor over decades and that is the common underlying theme of tumor genesis.

12. Cell birth and death in normal and timorous cells (20:53)

Just to go over that again in graphic form when a normal cell divides, every normal cell two cells are, of course, formed but one of those cells will die. So cell division you get two cells. One of those cells eventually dies leaving the net the same as the beginning one cell but in a tumor, it's quite different as, again, shown on this movie. The tumor cells, which are mutant we'll see in a few minutes why they're mutant the cell divides, but one of the daughter cells doesn't die. There will keep being more cells, more cells more cells over time and that starts a benign tumor.

13. Review of tumor definitions (21:43)

Again, to review the difference among tumors there are three kinds, all called tumors. The first is benign. It's not dangerous, it's not life-threatening and it's not invasive. The second type is malignant.

This is dangerous. It's locally invasive. That means it can invade surrounding tissues. Eventually, the malignant tumor may become metastatic in which case, it can travel to other parts of the body and that's when the real problem occurs. In the next chapter, we'll discuss the molecular basis for this process but now we'll quit and ask for questions.

14. Q&A: How can you tell if a benign tumor will become malignant? (22:35)

How much time do we have for questions? Five minutes. Good. Just raise your hand or light. Yes. How is it possible to tell if or when a benign tumor is going to turn malignant or metastatic? Can you all hear that question? The question was, how do you tell if a benign tumor is going to become malignant? The only person who can tell is the oracle. It's you can't. It's just stochastic. We'll see which means it's random. You can look at a benign tumor. You can estimate the probability that it may turn into a malignant tumor but you can't tell for sure and in the next chapter of this lecture I think you'll understand why. Other questions.

15. Q&A: Do cancer cells selectively metastasize to specific tissues? (23:36)

Yes. You said the cancer cells they travel through the blood vessels. Are they selective in the tissue they choose to Are the cancer cells selective for the tissues they choose? Yes, they are. Sometimes investigators call that "the seed and the soil." Certain kinds of cancers only grow in certain tissues in metastatic form. For example, prostate cancers generally go to the bone marrow whereas colon cancers generally go to the liver. Do we know the reason for that? The reason for that? No. We don't know the reason for that. In fact, we don't understand the basis for metastasis in general, either and that's what some of you are going to help us find out over the next ten years.

16. Q&A: Are malignant tumors harder to control than metastatic tumors? (24:20)

Q&A: Are malignant tumors harder to control than metastatic tumors? Yes. My name is Crystal Shavers, and I attend Margaret Murray Washington Career Senior High School. Is it more difficult to control a massive malignant tumor or a metastatic tumor? The question about controlling a massive malignant tumor or a metastatic tumor. Massive malignant tumors first of all, they usually don't become really massive as long as people have access to medical care because people notice them if they're really big but there are some tumors that have been taken out that have weighed 250 pounds. These are in morbidly obese people and they occur inside the abdomen and they thought they were just getting fat but they weren't. They had a tumor growing inside and that can be removed surgically. It's really not a problem until a metastasis because if the metastatic cells invade the liver or the lung you can't take out somebody's liver or lung so then the person is in trouble. So that's really the problem.

17. Q&A: Are malignant tumors harder to control than metastatic tumors? (25:30)

We have time for one more question. Yes. OK. Hi. Good morning. My name is Tiffany Fields. I attend Spingarn Senior High School and the question I wanted to ask was when you stated earlier that, like the tumors are caused when the cell keeps multiplying is there any way that we could, like paralyze that cell that's multiplying so that it won't be so many? Right. Good question. Can we paralyze cells so that they'll stop multiplying? In fact, that's what cancer therapy is is attempts to try and stop cancer cells from developing. There's also something called preventive drugs which try to prevent even early tumors from multiplying rather than shrinking tumors which are large. All cancer drugs do that now to some extent. Unfortunately, they also stop normal cells from growing so they're toxic. If any of you have ever seen patients who are treated for leukemia or cancer you know they lose their hair. They feel bad. They're nauseous. That's because the drugs, so far work on both normal cells and cancer cells. They're not selective. Hopefully, the next generation of drugs will be more selective. - OK. - Thank you. I'm not

going to throw them, but you can Can you hand them back there to those questioners?

18. Q&A: Can we paralyze cells so they stop multiplying? (27:01)

Thank you. All right. Let's go on to part two. Now, when I was treating Melissa back in the seventies for her leukemia we, again, had no idea of what caused her cancer. There were lots of theories. Theories are cheap. Some people thought that cancers were caused by viruses. Others thought that cancers were caused by bacteria or other infectious agents. Still others thought there was a breakdown in immunity. Most forms of cancer don't occur in people until they're middle-aged or older. It's also known that immunity tends to decrease with age so it was a reasonable theory that perhaps when immunity breaks down cancers pop up and still others thought that there were mutations in specific genes that cause tumors. Now, it turns out, in general that this last theory is the right one. That's the basis of the cancer revolution. Mutations in specific cancer genes cause the disease so cancer is, in essence, a genetic disease but it's different than most other genetic diseases with which you're familiar.

19. Theories on what causes cancer (28:26)

For example, let's look at some common genetic diseases. *DMD* is a gene which, when mutated causes muscular dystrophy. I assume you all know what muscular dystrophy is. It occurs in children, causes them muscle problems. It's a very severe disease. Now, some of you probably live in rural communities in Bowie or Gaithersburg. Anybody live there? Anyone live in an urban setting Washington, D.C., city? Now, the air is probably cleaner in the rural environment. You don't have all the car fumes. The environment may be better. If you had the same mutation in the *DMD* gene would that alter the disease that you would get? Would the muscular dystrophy be better or worse depending on where you live? Anybody? Who thinks it would be better if you lived in Gaithersburg? Who thinks anything would be better if you lived in Gaithersburg? Well, the truth of the matter is, of course it doesn't matter where you live. If you have a mutation in the *DMD* gene you're going to get exactly the same disease. It's totally independent of environment. The disease is only dependent on the specific mutation. A somewhat more complex case is caused by mutations in a gene called *LDLR* which predisposes to heart disease but not everybody who has a mutation in *LDLR* will get heart disease. It's not like muscular dystrophy where everyone who has a mutation in *DMD* will get muscular dystrophy. It's only if a person with an *LDLR* mutation is exposed to specific environmental influences in particular, diet, fats in the diet that they will get heart disease and the severity of the disease is then determined both by the genetic component this mutation that they inherit other genes, and their diet. So, it's more complex and cancer is even more complex than that although it's related. Some patients inherit a mutation in a gene which predisposes them to cancer. They won't necessarily get cancer but they are more likely to get cancer than a person who doesn't have an inheritable mutation in this gene and there are lots of genes. This is just an example of one of them *MLH1*. In order for them to actually develop a cancer they need more stuff, and in this case what they need is more mutations. No single mutation actually causes cancer. It's an accumulation of mutations in the same cell until the proverbial straw that breaks the camel's back. When enough mutations are accumulated then a cancer results and

20. Clonal expansion as a model of cancer (31:34)

you can think about this process in terms of waves of clonal expansion. Pretend each of these white balls represents a normal cell. Suppose one of those cells becomes mutant. Then it will proliferate. Its ratio of cell birth to cell death will increase to greater than one and it will proliferate over and above that of the normal cells. That is called a clone. Now, it's different than a clone like Dolly the sheep, right? That's an organism clone. This is just a cell clone, but it's the same idea. A cell with the same genetic characteristics is proliferating and making identical copies of itself but that's not the end of this story because once this mutant cell forms a benign tumor it can also get more mutations, or a cell from the

benign tumor can get another mutation and through a second wave of clonal expansion expand to become malignant, invasive and, of course the malignant cell or one of the malignant cells can subsequently accumulate another mutation and become metastatic. Any of you see that movie called *Multiplicity* with Michael Keaton? Well, it's a bit like that. Remember, Michael Keaton wanted to clone himself because he didn't have enough time. It turned out not to be such a great idea but he cloned himself. The first clone was OK, but the next clone was worse and with each successive clone there were more mistakes that were made that's like mutations till eventually, he got a very stupid Michael Keaton and it's the same kind of thing that happens in tumors successive waves of clonal mutations until you get a very bad one in this case, not stupid, but evil.

21. Cancer is usually not hereditary (33:40)

Now, it's important to recognize that these mutations like mutations that cause muscular dystrophy and heart disease are sometimes inherited, but usually they're not. Cancer, in general, is not a hereditary disease. There are cases of cancer which are hereditary as we'll discuss in a few minutes but generally the mutations occur after birth. Now, the only mutations that are hereditary are those that occur in egg or sperm cells. They can be transmitted from father or mother to son or daughter. Other mutations that occur in any of the other cells of the body obviously cannot be transmitted to a son or daughter. Now, the germ cells, the egg cells and the stem cells are only a very small proportion of the total cells the total trillion cells in the body 50 trillion cells. There are only a few million germ cells at any one time so it's just a really small fraction of germ cells. All the other mutations that occur in other cells aren't important. They're not hereditary, but they can cause cancers.

22. Types of genes that are mutated in cancer (34:58)

Now, what are these genes that get these mutations either inherited or after birth? There are only three kinds. One kind is called an oncogene. These genes normally because they're all normal until they're mutated everybody has them. Everybody has oncogenes, but they're normal and they normally stimulate growth. They're what you need to grow, to live but if an oncogene becomes mutated it's like having an accelerator in a car that's stuck to the floor because there's a heavy weight on the foot. The car continues to go even if the driver wants to stop it by lifting her foot off the accelerator pedal and that's just what a mutation in an oncogene does in a cell. It makes the cell continue to grow whereas normally, without the mutation the cell would stop growing because it's being controlled properly and this analogy works, too, for the second kind of gene which is called suppressor gene. These are the cell's brakes. These normally inhibit growth and a mutation in a suppressor gene is much like having a dysfunctional brake and just as cars have more than one brake they have a foot brake, they have a hand brake or emergency brake, and if all else fails you can pull the key out of the ignition cells have multiple brakes, too and it's only when several brakes plus an accelerator or two all become dysfunctional in an automobile that the car spins out of control and it's the same in a cell. It's only when several of these genes become mutated don't work properly, that the cell spins out of control and becomes a metastatic cancer. There's a third kind of gene which doesn't directly cause tumor genesis when mutant but only indirectly does it, and these are repair genes. Having a faulty repair gene is like having an inept mechanic can't fix the mistakes that are made and cells are always making mistakes always. Now,

23. Why has evolution not made DNA replication perfect? (37:20)

you'd think that with 5 billion years of evolution you'd have evolved a system that's not so prone to mistakes so we wouldn't have cancers through these mutations. Can anybody suggest why evolution hasn't gone that far and made DNA polymerases, *et cetera*, that are perfect? Yes. Because that would eliminate the variety in the genetic coding of that species. Mistakes are a part of evolution. In fact, you can look at cancer as a side effect of evolution. If mistakes weren't made, we'd all be amoebas. We

wouldn't be here in this auditorium. Unfortunately, cells are still making mistakes and they can cause disease both in hereditary diseases and cancer. Now, are our cells still making mistakes now? Christa. Somebody named Christa in the audience? Christa McAllister? OK. Look behind you. There's a guy named Dan. Is he a mutant? - No. - Look carefully! Look you sure? All right. Well, you're wrong. He's a mutant. We're all mutants, right? Because the egg and sperm that gave rise to us there was a mutation at least one, probably a few. Now, those mutations didn't occur in genes which cause disease, hopefully but we all are mutants in that sense.

24. Two main kinds of familial colon cancer: FAP and HNPCC (39:04)

Now, let's look for a minute about at these genes in more detail and we'll start with suppressor genes. Suppressor genes are important because they cause many forms of familial cancers inherited cancers. In the United States this year about 135,000 people will develop colon cancer in the world, about a million people this year. Most of them, eighty-five to ninety percent they're non-familial. That is, they'll be the only people in their family to develop colon cancer. No one else has their parents, their grandparents their siblings, their cousins so they're non-familial, but a small fraction, roughly ten to fifteen percent occur in a familial pattern and there are two major forms of familial colon cancer. One's called FAP, for polyposis and the other one is called HNPCC and the operative letters are "NP" for non-polyposis. A picture of a colon from a patient with polyposis is shown here. You can see all of those bumps are polyps. Polyps are just benign tumors. Sometimes they're called adenomas. That's why FAP actually stands for familial adenomas polyposis but we'll just call it polyposis for short and this patient has lots of these polyps. They're benign tumors, but there are lots of them. In fact, there are roughly 5,000 such benign tumors in the colon of each patient with polyposis. Some of those tumors eventually are going to progress to cancer. So patients, unless they're treated for this disease will generally get colon cancer by the time they're in their forties. They usually start to develop the polyps when they're in their teens. This syndrome is caused by a classic tumor suppressor gene called *APC*. It's just a cellular brake that goes awry can be inherited in a mutant form and cause this disease. The other form of hereditary colon cancer is caused by repair gene defects. That form is called HNPCC, non-polyposis and the difference between non-polyposis and polyposis is just what its name suggests. There are lots of polyps in polyposis but in HNPCC, the non-polyposis there's usually only a single tumor a single tumor shown here, a large tumor in fact, a cancer not thousands of polyps, just a single tumor and the genes that cause this disease are called mismatch repair genes. This animation shows how they work.

25. Animation: DNA mismatch repair (42:08)

There's a polymerase, DNA polymerase which copies both strands of the DNA the top strand and the bottom strand. Sometimes those two strands are called Watson and Crick strands but they're not perfect, as we just mentioned. Sometimes they make mistakes and the kind of mistake they might make is you'll see in a second to incorporate the wrong nucleotide. Normally, there's going to be an "A" opposite a "T" and a "C" opposite a "G" but suppose it makes a mistake and copies a "T" where a "C" should be? That should be "GC," but now there's a "T." So that's a mistake, a potential mutation. Fortunately, cells have repair systems that can erase those mutations and those repair proteins indicated here are called MSH2, MSH6, MLH1, PMS2. The names don't matter. What's important is that they recruit another enzyme called EXO1 exonuclease which chops off the mutant strand and then it allows a DNA polymerase to come by and synthesize the correct strand thereby fixing up the DNA and making it normal.

26. HNPCC can progress much faster than FAP (43:26)

Now, if a patient doesn't have a proper mismatch repair system then the cell with that defect will accumulate mutations much more quickly than a normal cell and that's why these patients with HNPCC develop cancer they have defects in this repair system and one of the things that's important about this

defect is it accelerates telescopes the whole process. In polyposis the whole scheme of mutations the multiple mutations in addition to the inherited ones that are required to get to a malignant or metastatic cancer generally takes twenty to thirty years so patients don't get tumors don't get malignant cancers or die from their disease until they're in their forties but in non-polyposis this whole process only takes a few years from a small, benign tumor to a malignant tumor only three to five years. The mismatch repair system in essence, functions like a spell checker on your word processing program.

27. Colorectal cancer pathway (44:38)

Now, let's put all this together and in colorectal tumors. Starting from normal cells the first mutation might cause a very small, microscopic small tumor, which then could grow over time to a large benign tumor. Both these are benign. Another mutation might cause a wave of clonal evolution causing a malignant tumor and a metastatic cancer. The genes that drive this process have been identified. *APC*, the gene that causes polyposis is the gene that starts it all off. It initiates the process. It's a cellular brake and then tumors enlarge when they acquire other mutations in oncogenes, like *K-Ras*. They become malignant when they acquire still other genes mutations in other genes like *Smad4* and *p53* and still mutations in other genes. An oncogene called *PRL-3* can help drive it to metastases. All these genes are mutated in most colon cancers.

28. p53 gene is mutated in most cancers (45:53)

Now, one of my favorite genes is *p53*. *p53* is mutated like all of the others in part because there's some sort of faulty repair but it's my favorite gene because a graduate student in our lab named Suzie Baker discovered its role about fifteen years ago and she was not much older than you guys, actually. She discovered that *p53* mutations occurred in just about all colon cancers and then another graduate student named Janice Nigro worked with her to find that *p53* was mutant in virtually every cancer that occurs throughout the world. It's hard to be a cancer without getting a *p53* mutation. So all of these different tumor types have mutations in *p53*. It's kind of a common denominator for cancer. And knowing that we can do some interesting kinds of experiments to try and relate the initial insult to the mutation and here are some examples: sunlight, such as you get when you're on the beach Everybody likes the beach. Anybody use a tanning salon? Raise your hand if you do. OK. Well, you not only will get a good tan. You may get a mutation at codon 2 at one of the residues of *p53* changes a "C" to a "T." Now, this is a characteristic mutation caused by sunlight. If you irradiate with ultraviolet light, sunlight bacteria, they get this kind of mutation a "C" to a "T" and that mutation occurs in the *p53* gene in skin cancers. Skin cancers are, of course, induced by ultraviolet light. You'd find them much more rarely in other forms of tumors which, of course, are not exposed to light. Another example is aflatoxin. It's a fungal toxin found in parts of Asia and Africa and it causes a different mutation at residue 249 of *p53*. It causes liver cancer so it's found only in the liver cancers of people who live in that area and are exposed to that toxin not found in other tumor types and of course, one of the worst forms of carcinogens are found in cigarette smoke. Cigarette smoke, we now know causes cancers by, in part, inducing mutations in *p53* at this position, and they, of course occur in lung tumors but not in other tumors and in colon cancers it's a more complex scenario. We know specific mutations in *p53* that occur but we don't know exactly what the carcinogen is. That's something that remains in the future. Now, how does *p53* work?

29. Animation: p53 regulates transcription (48:59)

p53 works...as shown in this animation. *p53* molecule, protein binds to specific sequences adjacent to genes which it controls. There's a *p53* molecule binding to its site, its binding site. It recruits an RNA polymerase, not a DNA polymerase that then makes RNA from these genes that it controls and the RNA is translated into two proteins. *p53* primarily regulates two proteins called, in the next slide, WAF and

PUMA.

30. Examples of proteins regulated by p53 (49:43)

Now, *WAF* is a cellular brake. *PUMA* is a different kind of gene. *WAF* one of the nice things about discovering genes is you can name them anything you want. *WAF* was named for Wafik El-Diery who was the student in our lab who discovered it and *PUMA* was named by Jen Yu because she's a runner and she liked their shoes. It's true. My favorite names for genes there's one in flower called *SUPERMAN* another one called *clark kent* so...one of the good things about being a scientist. At any rate, what do these genes do? The *p21* works as a brake. Look at this movie. Here's a typical brake that didn't work. If you don't have *p53* you don't have the regulation of *p21*, a brake. The *PUMA* works differently. It's a kind of a hara-kiri gene. If all else fails, what the cell does to control itself is to commit hara-kiri. Really, it's suicide. It's not accidental death. It's very purposeful. Scientific name is apoptosis but it's really a form of suicide and it protects the organism from the development of cancer. Obviously, it kills the cell, but it's good for the organism.

31. An example of an oncogene mutation in cancer (51:13)

And the last kind of gene which I'll just briefly touch because we'll discuss this further in the second lecture is oncogenes. Oncogenes, again are the accelerators in a car and oncogenes get activated often by chromosome translocations and in the second lecture we'll talk about one specific translocation that causes leukemia a translocation between chromosomes 9 and 22 and the interesting thing about this translocation is that its discovery has led to a novel therapy for that form of leukemia and that's where I'm going to stop and ask for questions.

32. Q&A: How do mutagens cause such specific mutations? (51:59)

Yes. How do mutagens produce such specificity in the mutations produced? How do mutagens produce such specificity? Mutagens are chemicals and chemicals bind to specific sequences within DNA and the proteins that DNA is packaged by. So, because they're chemicals like all chemicals, they have certain charges certain distributions of forces which direct them to specific molecules. Now, they'll react with lots of DNA throughout the cell. They only cause a problem if they mutate an oncogene or a suppressor gene. If they mutate a gene for hemoglobin, it doesn't matter. That's not going to cause a tumor but in an individual cell with a mutation of an oncogene or tumor suppressor gene, that could represent a step towards cancer.

33. Q&A: Is there a reason why mutagens change bases to thymine? (52:53)

Yes. My name is Craig Sweder. I'm representing Fairfax High School. In your chart that had *p53* with all the different cancers all of the base pairs were changed to thymine. Is there a reason for that, or is that just coincidence? I'll have to go back but I think it was coincidence. - OK. - Yes.

34. Q&A: Can you fix a mutated gene in hereditary cancer? (53:13)

I'm Ray Moratio of the Potomac School. If someone had a familiar cancer would there be any way to fix the mutated gene so that it couldn't be passed on? The question is, can you fix a mutated gene in an hereditary tumor type? And the answer is, in theory, yes in practice, no. You can do it. That's really not going to happen because of the serious ethical implications. What people can do is with *in vitro* fertilization techniques you can select an embryo cell that doesn't have a mutation to implant. So if a couple is known to have a hereditary mutation you can select an embryonic cell through *in vitro* fertilization that is devoid of the mutation thereby ensuring that particular child that develops from that

embryo will not have the disease but fixing it is not on the table.

35. Q&A: How would I distinguish my tumors as benign or malignant? (54:15)

Yes. I was just wondering, as a potential patient I have tumors all over my body, right and some of them are bigger than others but how would I distinguish between a benign and malignant and metastatic and when should I consult a doctor? That's a good question. The tumors you will generally see are the ones on your skin. If you feel anything inside you should immediately consult your doctor because you can't tell whether a tumor is benign or malignant from feeling it inside you but if you're talking about tumors on your skin a general rule is if it's round and circumscribed it's benign, and you don't have to worry about it especially if it's small. If it starts to grow, if you see it change or you see it start to spread or it turns color then you should immediately see a physician but everybody should see their physician once a year anyway and just ask him or her to look at any suspicious lesions. You shouldn't have to guess. That's what doctors are for to save you the trouble of making those kinds of decisions.

36. Q&A: Why aren't growing children affected more by cancer? (55:25)

Other questions. Yes. Why isn't cancer more prevalent amongst children who are, like who have rapidly dividing cells due to growth and That's a good question. Cancer is not simply a function of dividing cells. For example, cells of your small intestine divide much more frequently in fact, every day than the cells in your brain. Brain cells hardly ever divide yet brain tumors are pretty common and small intestinal cancers never occur. So it's something more than just cell division and we don't understand exactly what that is.

37. Q&A: Do cancer cells have multiple mutations in apoptosis factors? (56:02)

Yes. Lily Young, Thomas Jefferson High School. In cancer cells, are there usually multiple mutations in genes coding for apoptosis factors or is the mutation in *p53* enough to stop the process? Yeah. In every cancer, there forms In every cancer that forms there are several mutations, at least four or five and certainly in every metastatic cancer. So it's *p53*. In colon cancer, it's plus *APC*, plus *Ras*, plus other mutations in that one little site. All of them occur. Each one inactivates a different growth-controlling pathway. Again, it's very much like a car. You can control the car until you lose all your brakes and your accelerator. Losing just one, you can still control the car and that's a pretty good analogy for what happens in a cell. That's it. We have a break. See you in a minute.

38. Closing remarks by HHMI President Dr. Thomas Cech (57:07)

Thanks for the great talk, Bert, and now we're going to and I also thank the students for those fantastic questions. We're going to now take a thirty-minute break. When we return, Bert will continue his discussion of the genetics of cancer and how that's critical to both prevention and treatment.